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STUDIES ON THE TRANSMISSION AND VIRUS-VECTOR RELATIONSHIP OF RICE-TUNGRO VIRUS (RTV) IN DIFFERENT RICE GENOTYPE

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ABSTRACT

Rice is one of the most important cereal crops grown in India and West Bengal is the highest producing state covering more than 90% of the crop during Kharif season. There are various viral diseases which causes constraint in production of rice crop and are mostly transmitted by insects. Out of the 13 insect-borne rice viruses Rice tungro virus (RTV) is considered as one of the major diseases in rice. Rice tungro virus (RTV) in West Bengal are transmitted by different species of green leafhopper like Nephotettixvirescens, NephotettixnigropictusandZigzag leafhopper R. dorsalis. An experiment was conducted to study the role of these vectors in transmission of the RTV. In test tube inoculation method, highest percent of transmission on GLH was found on TN 1 (63.63) however in case of ZLH highest percent of transmission was found on IET4786 (47.36). However in case of cage inoculation method, both GLH (36.00) and ZLH (32.00) recorded the highest percentage transmission on TN1. Regarding virus vector relationship, the result obtained showed both the vectors transmitted the virus efficiently, but higher percentage of transmission was recorded with green leafhopper (GLH) and compared with ZLH. The retention period GLH was found upto 8 hours while the infectivity of ZLH was lost after 1 hour.

KEYWORDS: Rice Tungro Virus (RTV), Green leafhopper (GLH) and Compared with ZLH

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INTRODUCTION

Rice is one of the most important cereal crops grown all over the south East Asian countries. In India rice is grown in three seasons namely '*Aus*' (Pre-monsoon), '*Kharif*' or '*Aman*' (main crop in the monsoon season) and '*Boro*' (summer crop). West Bengal being the largest rice producing state in India covers more than 90% of the crop during *Kharif* season with high yielding variety (HYV) where as in *boro* season all the cultivated lands are covered with HYV. Cultivation of *boro* rice depends on availability of irrigation water. Rice crop (*Oryzasativa*) is infected by at least 13 virus diseases and they are mostly transmitted by insects. Out of the 13 insect-borne rice viruses, Rice tungro virus (RTV) is considered as one of the major diseases in rice growing countries of the south East Asia. This disease caused severe outbreak periodically in many rice growing countries including India. In India, RTV is most predominant in *kharif* season. The major outbreak of RTV has been noticed from different countries of south east Asia that include Philippnes, Thailand, India, Malaysia, Bangladesh, Srilanka and many other countries (Anjaneyulu*et al.,* 1994). Although the detailed information on the estimate of crop loss from India is not presently available, but from West Bengal Chowdhury (1997) reported an estimated crop loss which exceeds more than 10 crore due to the wide spread incidence of RTV in various districts of the state. The most important vector for RTV under West Bengal situation are different species of green leafhopper like *Nephotettixvirescens* and *Nephotettixnigropictus*

(Mukhopadhyay and Chowdhury, 1970). Population of rice green leafhopper is abundant during kharif season which appears from the month of July and increased gradually in September to October and declined from the beginning of December. In extreme cold and in hot summer, they are almost absent (Mallik and Chowdhury, 2000). Considering the importance of RTV in West Bengal, this study was undertaken to find out the role of *R. dorsalis and Nephotettixvirescens* in transmission of RTV.

MATERIALS AND METHODS

The present experiment was conducted at Regional Research Station (OAZ), UBKV, Majhian, DakshinDinajpur West Bengal 2014-15 using six rice varieties viz., IR – 36, IR – 62, IR – 64, IET 1444, IET 4786 and TN1 (Taichung Native 1).

Collection and Maintenance of Virus Source Plant

Rice plant having typical tungro symptoms were collected from field and were planted in pots containing well pulverised field soil with organic matter and required quantity of inorganic fertilizer. After that potted RTV infected plants were watered and kept under insect proof condition for its establishment. Dried leaves were removed from the source plant before they were used for acquisition of virus.

Monitoring of Vector

Both zigzag leafhoppers (*Reciliadorsalis*) and green leafhoppers (*Nephotettixvirescens*) were monitored and collected from rice seed beds and transplanted rice with the help of conical shaped sweeping net, made with fine nylon net of 30 cm diameter following the usual sweeping techniques. Leafhoppers (both types) caught in each sweep were sorted out with the help of aspirator and counted. The collected leafhoppers were placed in glass tube with rice plants and brought to the laboratory for rearing.

Vector Rearing in Cage

Both rice green leafhoppers (GLH) and zigzag leafhoppers (ZLH) collected from rice fields were released on healthy rice seedlings for 24 hrs. To make them virus free. After making the leafhoppers (both types) non viruliferous, they were transferred to rearing cages made up of wood and nylon net with dimensions of 3 ft. x 1.5 ft. x 1.5 ft. For rearing of leafhoppers, 35 – 40 days old healthy TN1 (Taichung Native 1) seedlings @ 5 seedlings/pot were taken and raised in an earthen pots. Sufficient number of GLH and ZLH collected previously were then separated and transferred to the rearing cage. Rice plants were replaced by fresh plants after 5 days of interval and the replaced old plants were placed in separate cages for development of fresh insect from eggs hatched in the rice plants. The nymphs that emerged from the eggs were allowed to grow for adult and they were used for transmission studies.

Raising of Seedlings for Transmission Studies

For transmission study, one-week-old rice seedlings of different varieties viz., IR – 36, IR – 62, IR – 64, IET 1444, IET 4786 and TN1 (Taichung Native 1) raised in earthen pots were used for test tube or cage inoculation. Such seedlings were uprooted from the pots washed with tap water and transplanted in small pots for mass inoculation or used for test-tube inoculation.

Transmission Test

Transmission efficiency of Rice tungro virus (RTV) using the vectors rice green leafhopper (*Nephotettixvirescens*) and zigzag leafhopper (*Reciliadorsalis*) was studied in 6 varieties commonly grown in West Bengal under two condition i.e. (i) using single insect in one seedling (test-tube inoculation) and (ii) mass inoculation method (Cage method).

Test Tube Inoculation Method

RTV infected rice plants maintained in the laboratory were used as source plants for virus transmission. Sufficient number of both fresh zigzag leafhoppers and green leafhoppers were collected separately from the rearing cages with the help of aspirator and released in the cage having RTV infected plants for 24 hrs. acquisition feeding period. After completion of acquisition feeding period on the source plants, the viruliferous insects were taken out with the help of aspirator and released on Ten (10) days old healthy seedling of test varieties previously placed individually in test tube of ½ inch.diameter x 6 inch. length containing little amount of water to keep moist the root zone of the seedlings. In each tube one insect that aquired RTV from source plants were released by aspirator. Top of the test tube were covered with piece of markin cloth and tied with rubber bands to protect the insect from escaping from the test tubes. The test tubes were kept in test tube rack. After 24 hrs.of inoculation feeding, insects were taken out and the inoculated seedlings were transplanted in earthen pots previously filled with paddy field soil. The inoculated seedlings were sprayed with an insecticide (0.1% metasystox) and kept in insect proof condition for symptom development.

Cage Method of Virus Inoculation

In this method large number of seedlings was exposed to viruliferous insect to test transmission efficiency of vector in different varieties. The RTV infected plants maintained in the laboratory were kept in the cage, and both fresh zigzag leafhoppers and green leafhoppers were collected from rearing cages with the help of aspirator and released in the cages for 24 hrs. For acquisition feeding. After competition of 24 hrs. of acquisition feeding the viruliferous insects were taken out with the help of aspirator and released in cage having healthy potted seedlings (3–4 GLH/seedling) of different rice varieties for inoculation feeding. After 24 hrs.of inoculation feeding potted seedlings were taken out and kept in insect proof condition for symptoms development and sprayed periodically with an insecticide. Transmission efficiency of the vector was judged after development of tungro symptoms which usually appeared 15 days after inoculation.

Virus Vector Relationship

Virus-vector relationship of the two types of vector was also determined by considering acquisition feeding time, inoculation feeding time and retention period in the vector. To determine the virus-vector relationship most susceptible variety TN1 was used.

RESULTS

Comparison on the Transmission Efficiency of Rice Tungro Virus (Rtv) by Green Leafhopper (Glh) and Zigzag Leafhopper (Zlh)

The results of test-tube inoculation on the rate of virus transmission are presented in table 1.It was found that both the vectors efficiently transmitted the tungro virus irrespective of the varieties. In test-tube inoculation method of GLH the highest percentage of transmission was recorded in TN1 (63.63) followed by IET1444 (46.66), IET4786 (45.45), IR64 (44.44), IR36 (36.84) and IR62 (26.66) respectively. However, when ZLH was inoculated in test-tube IET4786 (47.36)

recorded the highest percent of transmission followed by TN1 (45.45), IR64 (35.29), IR 36 (33.33) and IR62 (25.00) respectively. Results in the cage inoculation (mass inoculation) method are presented in table 2 showed slightly higher percentage of transmission with GLH than the ZLH. In this case with GLH highest percentage of transmission was recorded in TN1(36.00) followed by IET1444 (24.00), IR62 (20.00), IR64 (20.00), IR36 (16.00) and IET4786 (12.00) respectively. Similarly, in ZLH inoculated cage the highest transmission was recorded on TN1(32.00) followed by IET1444 (20.00), IR64 (16.00), IR62 (12.00), , IR36 (12.00) and IET4786 (12.00) respectively.

Virus-Vector Relationship

The results regarding virus vector relationship are presented in table 3 and 4. It is evident from the result that both the vectors transmitted the virus efficiently but higher percentage of transmission was recorded with green leafhopper (GLH) and compared with ZLH. After 30 minutes' acquisition time transmission percentage with GLH was found higher (26.66%) than ZLH (6.66%). Upto 1 hour acquisition time trend of transmission percentage in case of GLH was also found higher than ZLH and after that it gradually increased on both the cases. At higher period of acquisition feeding time from 2, 3 and 4 hours the rate of transmission in case of GLH were 53.33, 46.66 and 53.33 percent respectively while in case of ZLH on the same time of acquisition, percentage of transmission were 33.33, 40.00 and 46.66 percent respectively. However, no transmission was observed with ZLH when given 30 minutes of inoculation feeding time. At 1 hour and 2 hours of inoculation feeding time transmission percentage recorded were 20% in both the cases. Exposing 3 and 4 hours of acquition feeding time percentage of transmission by ZLH was found to increase but increasing the incubation period beyond 1 to 3 hrs. recorded almost same percentage of transmission. Based on the aquistion and inoculation feeding it could be concluded that transmission efficiency of GLH was more than the ZLH. When the retention period of the virus in the vector was taken into consideration, it showed that GLH retained the virus upto 8 hours while the infectivity of ZLH was lost after 1 hour (Table 4).

DISCUSSIONS

Transmission of the tungro virus by the genus *Nephotettix* has been investigated in detailed by different workers and suggested that *Nephotettixvirescens* is the major vector of tungro virus. Transmission of RTV by its vector is of semi-persistent type and the vector could acquire and inoculate the virus within a short feeding and inoculation time (Hibino *et al.*, 1977). Further tungro is a complex virus consisting of rice tungro bacilliform virus (RTBV) and rice tungro spherical virus (RTSV) and both of them are transmitted in semi-persistent manner. Rivera *et al.* (1969, 1972) reported that only 6-8% transmission by *R. dorsalis* but the present studies indicated a higher percentage of transmission with this vector. The virus-vector relationship of tungro virus with its vector *N. virescens*has been studied critically by various workers at different places (Ling, 1966, 1972; Mukhopadhyay and Chowdhury, 1973). However, Hibino *et al.* (1979) and Chowdhury *et al.* (1990) observed a maximum retention of RTBV upto 5-6 days by leafhopper (*N. virescens)*. Futhermore, they also observed RTSV has a shorter retention period than RTVB. Rivera and Ou, 1965 reported that the shortest acquisition access period for *N. virescens*was 30 minutes and there was no latent period in the vector and could transmit immediately after acquisition feeding on virus source plant.

CONCLUSIONS

It may be concluded that Green leaf hopper (GLH) is the potent vector for transmission of rice tungro virus and also has the highest retention period as compared to Zigzag leaf hopper (ZLH). Even in minor form, rice tungro virus

(RTV) has also been seen in many of the important varieties grown in West Bengal, therefore a good knowledge for better management of the disease is in need of an hour so that the losses can be avoided and the economy of the farmers remains unaffected.

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APPENDICES

Table 1: Comparison on the Transmission Efficiency of Rice Tungro Virus (RTV) by Green Leafhopper and Zigzag Leaf Hopper on Some Rice Varieties by Test Tube Inoculation using Single Insect

Variety	Percentage of Transmission					
	Green Leafhopper (GLH)			Zigzag Leafhopper (ZLH)		
	No. of Plant Inoculated	No. of Plant Developed Symptoms	Percentage of Transmission	No. of Plant Inoculated	No. of Plant Developed Symptoms	Percentage of Transmission
IR – 36	19	7	36.84	15	5	33.33
IR – 62	15	4	26.66	16	4	25.00
IR – 64	18	8	44.44	17	6	35.29
IET – 1444	15	7	46.66	16	5	31.25
IET – 4786	22	10	45.45	19	9	47.36
TN1	20	14	63.63	22	10	45.45

Table 2: Comparison on the Transmission Efficiency of Rice Tungro Virus (RTV) by Green Leafhopper and Zigzag Leaf Hopper on Some Rice Varieties by Mass Inoculation using Caged Method

Variety	Percentage of Transmission					
	Green Leafhopper (GLH)			Zigzag Leafhopper (ZLH)		
	No. of Plant Inoculated	No. of Plant Developed Symptoms	Percentage	No. of Plant Inoculated	No. of Plant Developed Symptoms	Percentage
IR – 36	25	4	16.00	25	3	12.00
IR – 62	25	5	20.00	25	3	12.00
IR – 64	25	5	20.00	25	4	16.00
Table 2: Contd.,						
IET – 1444	25	3	12.00	25	3	12.00
IET – 4786	25	6	24.00	25	5	20.00
TN1	25	9	36.00	25	8	32.00

Table 3: Comparison on the Virus-Vector Relationship of Rice Tungro Virus (RTV) by Nephotettixvirescens and Recilia Dorsalis in TN1 Rice Variety

Character	Transmission Percentage					
	Green Leafhopper (GLH)			Zigzag Leafhopper (ZLH)		
	No. of Plant Inoculated	No. of Plant Developed Symptoms	Percentage	No. of Plant Inoculated	No. of Plant Developed Symptoms	Percentage
Acquisition Feeding Time 30 min.	15	4	26.66	15	1	6.66
1.0 hr.	15	7	46.66	15	2	13.33
2.0 hr.	15	8	53.33	15	5	33.33
3.0 hr.	15	7	46.66	15	6	40.00
4 hr.	15	8	53.33	15	7	46.66
Inoculation Feeding Time 30 min.	15	3	20.00	15	0	0
1.0 hr.	15	6	40.00	15	3	20.00
2.0 hr.	15	7	46.66	15	3	20.00
3.0 hr.	15	7	46.66	15	4	20.66
4.0 hr.	15	7	46.66	15	4	26.66

Table 4: Retention Period of Virus in the Vectors *Nephotettixvirescens*and Recilicdorsalisafter Acquition of Virus from the Source Plant

Vectors	Retention Periods after Different Period of Acquisition (in Hr.)			
	1 Hr.	3 Hrs.	6 Hrs.	8 Hrs.
Nephotettixvirescens	+	+	+	+
Reciliadorsalis	+	-	-	-